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## Growth Responses of Blueberry Softwood Cuttings to Atmospheric Carbon Dioxide Enrichment

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### Summary

Softwood cuttings of 'Earlyblue', 'Spartan', 'Blueray', and 'Bluecrop' cultivars of highbush blueberry (*Vaccinium corymbosum* L.) and those of the wild species Oobasunoki (*Vaccinium smallii*) were planted in a moisture peat moss medium and were grown in growth chambers at ambient (current atmospheric concentration) or elevated (1,000 mg l<sup>-1</sup>) levels of carbon dioxide from the July 16 to November 14, 2002. The experiment was finished on April 8, 2003. The carbon dioxide enrichment had a definite positive effect on the growth of blueberry softwood cuttings. The effect was manifested in improved rooting and surviving ratios, earlier root induction, longer roots, and increased branch growth. It was concluded that carbon dioxide application is a feasible means for achieving faster and efficient propagation of highbush blueberry softwood cuttings.

**Key words** : CO<sub>2</sub> enrichment, highbush blueberry, propagation, softwood cuttings

### Introduction

Rising atmospheric carbon dioxide concentration has become a global problem (Hsiao and Jackson, 1999). In 1998, the concentration of atmospheric carbon dioxide gas in the air reached the level of 368 mg l<sup>-1</sup> (Mearns, 2000). The effects of the increased carbon dioxide concentration on growth of rice plants and other agronomic and forest species are well documented (Lawlor *et al.* 1991, Kimball *et al.* 2002). Sionit *et al.* (1980) reported that semi-dwarf spring wheat under well-watered conditions showed a 43% increase in the rate of tiller production and significant increases in grain yield, total dry matter, and number and size of the grains in response to atmospheric carbon dioxide enrichment. Sionit (1983) also reported that the limiting effect of nutrient supply could be partly overcome by an increase in the atmospheric carbon dioxide concentration, as was shown in the case of soybeans. Young bean plants were found to grow faster in air enriched with carbon dioxide than in ambient carbon dioxide (Porter and

Grodzinski, 1983). Also, the leaf area and dry weight of the bean plants and the starch content in the leaves and stem were significantly increased by carbon dioxide enrichment. Leaf thickness also responded positively to carbon dioxide enrichment (Radoglou and Jarvis, 1992). Scheidegger and Nosberger (1984) found that elevated carbon dioxide concentration positively influenced the dry weight and the starch and sugar accumulation of white clover plants. Rogers *et al.* (1983) reported that corn, soybeans, loblolly pine, and sweetgum species responded with increased yield and wood volume. Water-stressed sweetgum and loblolly pine plants grown in air with elevated carbon dioxide concentration were found to have greater total dry weight (Tolley and Strain, 1984). Tissue *et al.* (1993) found that photosynthetic rates were higher for loblolly pine plants grown at elevated carbon dioxide only when they received supplemental nitrogen. Carbon dioxide enrichment increased the total dry weight by an average of 38% and increased the number, length, diameter and specific weight of the needles of Monterey pine seedlings in studies by Conroy *et al.* (1986, 1987). Carbon dioxide enrichment was reported to enhance the growth of white oak seedlings by 85%, with the greatest enhancement being in the root system, and to increase the proliferation of fine roots (Norby *et al.* 1986). Faria *et al.* (1996) reported that the leaves of cork oak grown in a carbon dioxide-enriched atmosphere were protected from the short-term effects of high temperature and that plants grown in an atmosphere with elevated carbon dioxide concentration had positive net carbon uptake rates during a heat shock treatment. They also reported that their recovery after heat shock treatment was faster than that of plants grown in ambient conditions. Radoglou and Jarvis (1990 a, b) reported that four poplar clones responded positively to elevated carbon dioxide with increases in number of leaves, total length of stem, total leaf area, overall growth rate, and total leaf, stem and root dry weights. On the other hand, Norby and O'Neill (1990) found that an increase in dry weight of yellow poplar seedlings, resulting from elevated carbon dioxide, occurred only in root systems. The increase in total dry weight of carbon dioxide-enriched sweet chestnut seedlings was reported to be the result of an increase in root dry weight (Mousseau and Enoch, 1989). Glenn and Welker (1997) found that increased root carbon dioxide resulted in increased root growth without increase in the shoot growth of peach trees in soil and hydroponic systems.

Highbush blueberry (*Vaccinium corymbosum* L.) propagation using softwood cuttings is less successful than that of rabbiteye blueberry (*V. ashei* Reade) because of the rooting difficulty of highbush blueberry (Gough, 1994). On the other hand, including the chilling period, hardwood cuttings of highbush blueberry need a longer growing time than that required for the softwood cuttings (Gough, 1994). Therefore, the aims of this study were to establish methods to improve the rooting efficiency of softwood cuttings by elevated carbon dioxide

levels and to shorten the nursery time of blueberry clones.

### Materials and methods

A preliminary experiment was carried out from July to April in 2002, and a similar experiment was carried out from July 16 to April 8 in 2003. The two experiments were conducted under similar conditions. Five cultivars were used in both experiments: 'Earlyblue', 'Spartan', 'Collins', 'Blueray', and 'Patriot' were used in the first experiment, and 'Earlyblue', 'Spartan', 'Blueray', and 'Bluecrop' cultivars of highbush and Oobasunoki (*Vaccinium smallii*), a wild species blueberry growing near the Field Science Center of Tohoku University in Naruko, were used in the second experiment. Fifty softwood cuttings of each cultivar and 30 softwood cuttings of Oobasunoki were inserted into moisture peat-moss media in rooting boxes, and the boxes were placed in growth chambers under a carbon dioxide-enriched (Fig. 1.) or an ambient (Fig. 2.) atmosphere. The carbon dioxide concentration in the enriched atmosphere chamber was about three-times higher than that in the ambient one (1,000 ppm on average). These conditions were maintained during the summer and autumn periods until the falling of leaves. In the second experiment, the first sampling, 20 of the 50

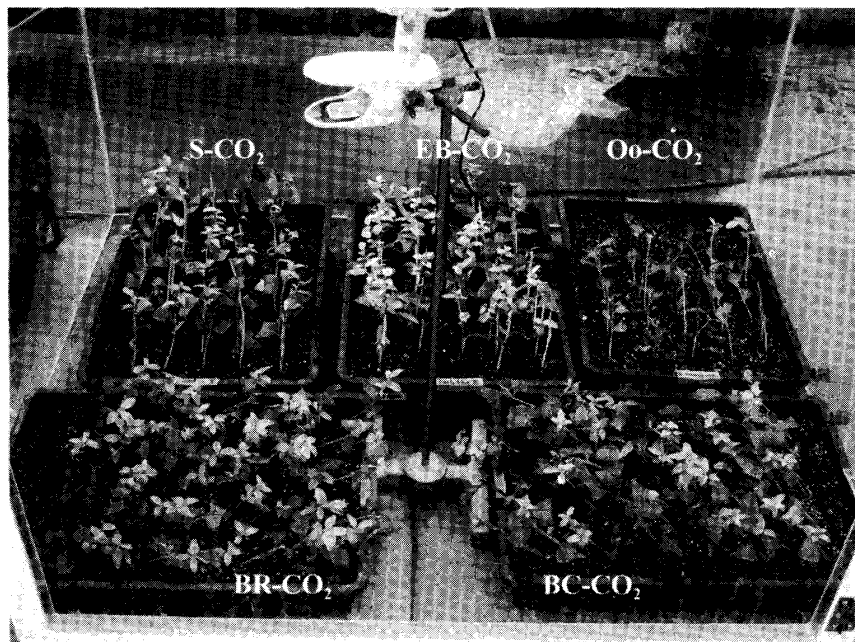


FIG. 1. Growth of blueberry softwood cuttings in a carbon dioxide-enriched atmosphere (August 2002).

S-CO<sub>2</sub>: Spartan, carbon dioxide treatment; EB-CO<sub>2</sub>: Earlyblue, carbon dioxide treatment; Oo-CO<sub>2</sub>: Oobasunoki, carbon dioxide treatment; BR-CO<sub>2</sub>: Blueray, carbon dioxide treatment; BC-CO<sub>2</sub>: Bluecrop, carbon dioxide treatment.

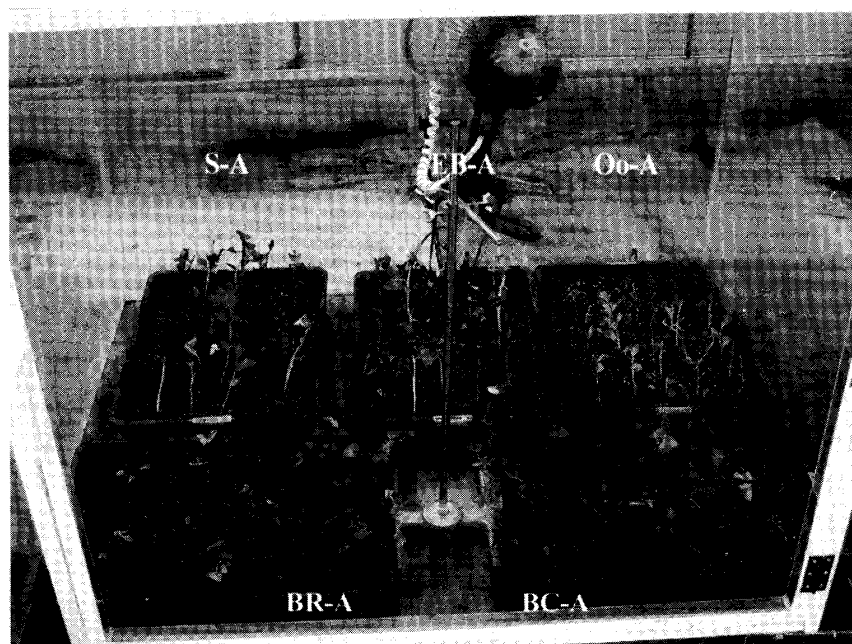


FIG. 2. Growth of blueberry softwood cuttings in an ambient atmosphere (August 2002).

S-A : Spartan, ambient treatment ; EB-A : Earlyblue, ambient treatment ; Oo-A : Oobasunoki, ambient treatment ; BR-A : Blueray, ambient treatment ; BC-A : Bluecrop, ambient treatment.

cuttings (10 of the 30 cuttings for Oobasunoki), was done on November 14, 2002. At the first sampling, both the number of rooted cuttings and the number of living cuttings were counted and the lengths of the roots were also measured. The rooting ratio and the survival ratio of each cultivar were calculated, and the ratios of cultivars grown in the carbon dioxide-enriched chamber and the ratios of those grown in the ambient chamber were compared. The second sampling was done after finishing the experiment in April. At this time all of the previous indexes were recorded, and the growth rates of new branches were measured. Only the data obtained in the second experiment are shown in this report because the results of the two experiments were similar.

## Results and discussion

### *Number of rooted cuttings and rooting ratio*

The softwood cuttings of blueberry treated at a higher carbon dioxide concentration showed a greater number of rooted cuttings (data not shown) and also a higher rooting ratio than those of the ambient samples as shown in Fig. 3. The highest rooting ratio was in 'Bluecrop' and the lowest ratio was in the 'Spartan' cultivar grown in the carbon dioxide-enriched atmosphere. The 'Spartan' cultivar also had the lowest rooting ratio in the ambient treatment, while the

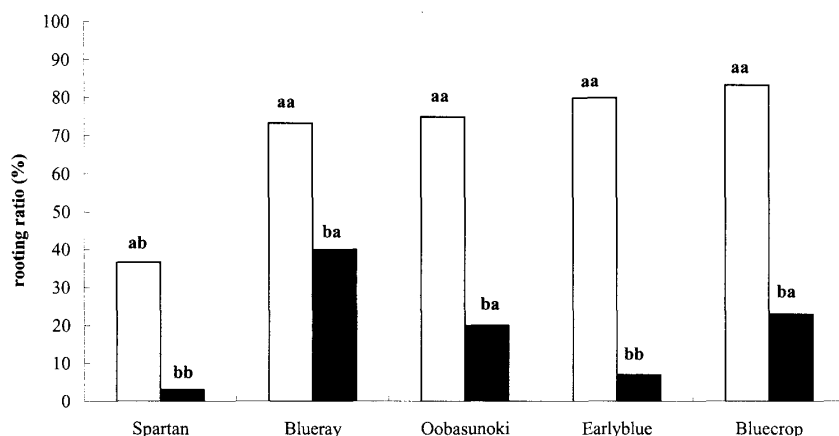


FIG. 3. Rooting ratios of blueberry softwood cuttings at the second sampling.

□ Elevated carbon dioxide treatment, ■ Ambient treatment.

Means within the same treatment and within the same cultivars were compared by Tukey's HSD test at the 0.05 level of probability.

ab, bb, bab, etc...: the first letter shows significant difference between the treatments, and the second and third letters show the significant differences between cultivars.

highest ratio was in 'Blueray'. The difference between rooting ratios of the carbon dioxide-treated and ambient plants, was also observed among the cultivars. 'Earlyblue' and 'Spartan' cuttings in the carbon dioxide treatment group showed rooting ratios about 10-times higher than those in the ambient group, while 'Blueray' and Oobasunoki cuttings showed only 2- and 3-times higher rooting ratios, respectively, in the carbon dioxide-enriched atmosphere. 'Bluecrop' had a 4-times higher rooting ratio in the carbon dioxide treatment chamber than that in the ambient chamber.

#### *Number of living cuttings and survival ratio*

There was little difference among the cultivars in number of living cuttings (data not shown) and survival ratio in the carbon dioxide-enriched atmosphere as shown in Fig. 4. The survival ratios of 'Blueray' and 'Bluecrop' were the highest, while that of 'Spartan' was the lowest. The survival ratio of 'Blueray' cuttings grown in the ambient atmosphere was the highest among the plants tested, while the ratios of 'Spartan' and 'Earlyblue' cuttings were the lowest. The survival ratios in the elevated carbon dioxide atmosphere were about 11-times higher in 'Earlyblue' cuttings, 10-times higher in 'Spartan' cuttings and about 4-times higher in 'Bluecrop' cuttings than those of cuttings in the ambient atmosphere.

#### *Root growth*

The lengths of roots were measured and recorded at the first sampling and the second sampling (Fig. 5). A comparison of root lengths at the first and second

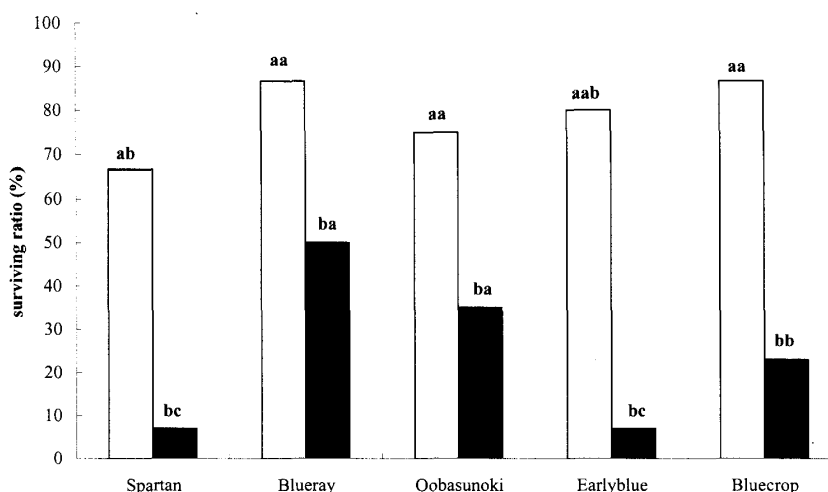


FIG. 4. Surviving ratios of blueberry softwood cuttings at the second sampling.   
 □ Elevated carbon dioxide treatment, ■ Ambient treatment.   
 Means within the same treatment and within the same cultivars were compared by Tukey's HSD test at the 0.05 level of probability.   
 ab, bb, aab, etc... : see Fig. 3.

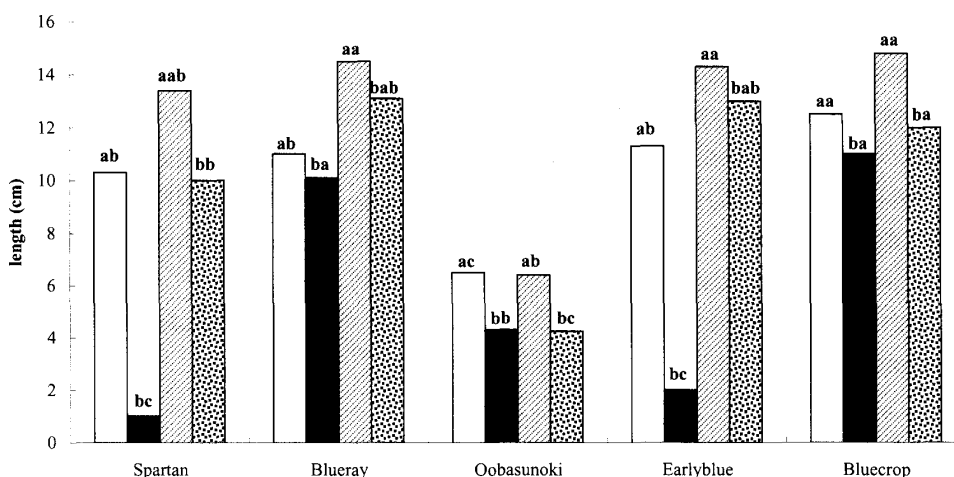


FIG. 5. Root lengths of blueberry softwood cuttings at the first and second sampling.   
 □ Elevated carbon dioxide treatment at the first sampling, ■ Ambient treatment at the first sampling, ▨ Elevated carbon dioxide treatment at the second sampling, ▩ Ambient treatment at the second sampling.   
 Means within the same treatment and within the same cultivars were compared by Tukey's HSD test at the 0.05 level of probability.   
 ab, bb, aab, etc... : see Fig. 3.

samplings would clarify the effect of elevated carbon dioxide on the growth rate of softwood cuttings. The cuttings of all the cultivars grown in the elevated carbon dioxide atmosphere had longer roots than those grown in the ambient atmosphere. Carbon dioxide treatment seemed to be the most effective for early root induction of 'Spartan' and 'Earlyblue' cuttings compared to the other

cultivars. They had about 5-times longer roots in the early stage in the elevated carbon dioxide atmosphere than those in the ambient atmosphere. Among the cultivars grown in the elevated carbon dioxide atmosphere, the 'Bluecrop' cultivar had the longest roots at both the first and second samplings, while the Oobasunoki wild species had the shortest ones. In the case of ambient treatment, the 'Bluecrop' had the longest roots at the first sampling and 'Blueray' had the longest roots at the second sampling, whereas 'Spartan' had the shortest roots at the first sampling and Oobasunoki had the shortest roots at the second sampling.

#### *Growth of the new branches*

The growth of new branches was only measured at the second (spring) sampling, and the data are shown in Fig. 6. Carbon dioxide treatment had a clear effect on growth increment but the growth of new branches was much more influenced by the cultivar effect. Among the carbon dioxide-treated cultivars, the 'Bluecrop' cultivar showed the longest new branches and Oobasunoki showed the shortest new branches. In the ambient cuttings, the longest branches were also found in the 'Bluecrop' cultivar and the shortest ones were found in the 'Earlyblue' cuttings. The greatest effect of the elevated carbon dioxide level on new branch growth was found in the 'Earlyblue' cuttings, the branch growth being two-times greater than that of ambient ones. The least effect of carbon dioxide enrichment on new branch growth was observed in the Oobasunoki cuttings.

The results showed that, carbon dioxide enrichment had a definite positive effect on the growth of softwood cuttings of both blueberry cultivars and the wild species. The effects were manifested in improved rooting ratio and survival ratio,

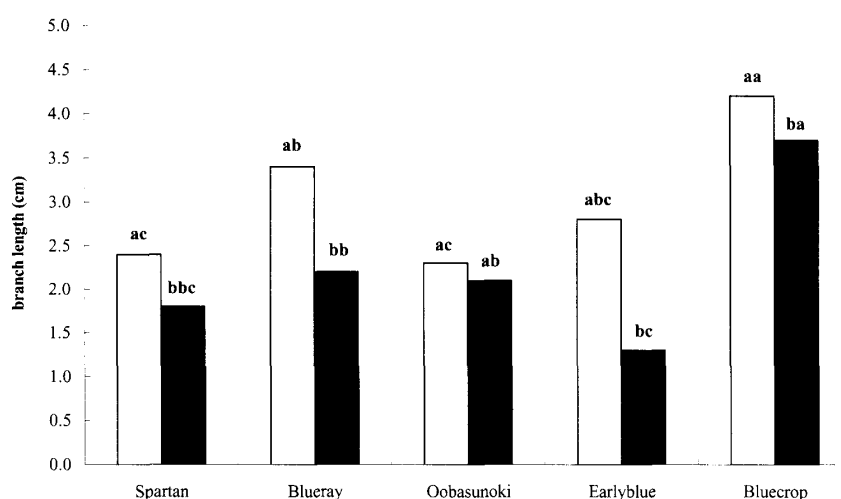


FIG. 6. Growth of new branches at the second sampling.

□ Elevated carbon dioxide treatment, ■ Ambient treatment.

Means within the same treatment and within the same cultivars were compared by Tukey's HSD test at the 0.05 level of probability.

ab, bb, aab, etc... : see Fig. 3



earlier root induction, longer roots, and increased growth. Although the responses of the cultivars to atmospheric carbon dioxide enrichment were different, all of the cultivars responded much more strongly than did the wild species, Oobasunoki. Finally, it can be concluded that carbon dioxide application is a feasible means for achieving faster and efficient propagation of highbush blueberry softwood cuttings.

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